

## PREVALENCE OF ANTIBIOTIC-RESISTANT STAPHYLOCOCCUS AUREUS STRAINS IN HOSPITAL ENVIRONMENTS AND ASSOCIATED INFECTION RISKS

*Original Article*

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## Abstract

**Background:** Antibiotic-resistant *Staphylococcus aureus* (ARSA), particularly methicillin-resistant *S. aureus* (MRSA), remains a critical cause of healthcare-associated infections worldwide. Its ability to persist on hospital surfaces and spread among patients and healthcare workers poses a considerable public wellbeing threat. Understanding the environmental distribution and resistance patterns of *S. aureus* is essential for helpful infection control strategies.

**Objective:** To determine the prevalence and distribution of antibiotic-resistant *Staphylococcus aureus* strains in hospital environments and assess their correlation with infection incidence among patients and healthcare workers.

**Methods:** This cross-sectional analysis was done over five months in three tertiary care hospitals in KPK, Pakistan. A total of 350 samples were collected from environmental surfaces (n=200), patients (n=100), and healthcare workers (n=50). Isolation and identification of *S. aureus* were completed via typical microbiological procedures. Antibiotic exposure was tested by the Kirby–Bauer disk diffusion way tracking CLSI guidelines. Details were evaluated using SPSS version 27, applying Pearson’s correlation and logistic regression tests ( $p < 0.05$  considered significant).

**Results:** Out of 350 samples, *S. aureus* was isolated from 156 (44.6%), of which 87 (55.8%) were MRSA. The highest contamination occurred in ICUs (62.3%), followed by surgical wards (54.7%). Environmental MRSA prevalence showed a strong positive correlation with patient infection incidence ( $r = 0.72$ ,  $p < 0.001$ ). All isolates were resistant to penicillin (94.2%) but remained fully sensitive to vancomycin and linezolid.

**Conclusion:** Antibiotic-resistant *S. aureus* is highly prevalent in hospital environments, with significant links to patient infections. Strengthened environmental cleaning, healthcare worker screening, and antibiotic stewardship are vital to curb transmission and improve infection control outcomes.

**Keywords:** Antimicrobial Resistance, Cross-Sectional Studies, Environmental Monitoring, Hospital-Acquired Infections, Methicillin-Resistant *Staphylococcus aureus*, Microbial Sensitivity Tests, Staphylococcal Infections

## Introduction

*Staphylococcus aureus* remains one of the most significant pathogens in both community and hospital settings, representing a leading cause of a wide range of infections that vary from mild skin conditions to life-threatening systemic diseases (1). In hospital environments, its ability to persist on surfaces and medical devices, coupled with its capacity for acquiring resistance to multiple antibiotics, poses a serious challenge to infection control and patient safety (2). Among the resistant strains, methicillin-resistant *Staphylococcus aureus* (MRSA) has become a particularly alarming public health concern due to its high morbidity, mortality, and the limited therapeutic options available for treatment (3). Despite rigorous infection prevention measures, the persistence and dissemination of antibiotic-resistant *S. aureus* in healthcare facilities continue to endanger both patients and healthcare workers, underscoring the urgent need for updated surveillance and research into its prevalence and impact (4). Antibiotic resistance in *S. aureus* has evolved over decades of antibiotic exposure and misuse (5). The initial success of penicillin in the 1940s was soon undermined by the emergence of penicillin-resistant strains, followed by methicillin resistance identified in the early 1960s (6). Today, MRSA is not confined to clinical settings but also circulates in communities, livestock, and even the environment, reflecting the adaptive potential of this bacterium. In hospitals, where antibiotic pressure and susceptible hosts coexist, resistant *S. aureus* strains have found an ideal niche for survival and transmission. Environmental contamination—ranging from bed rails and medical instruments to healthcare workers' hands—serves as a critical reservoir facilitating cross-transmission (7). Such contamination, when not properly managed, contributes substantially to hospital-acquired infections (HAIs), increasing patient morbidity, prolonging hospital stays, and escalating healthcare costs. The hospital environment plays a crucial intermediary role in the epidemiology of *S. aureus* infections (8). Numerous studies have demonstrated that resistant strains can survive for extended periods on dry surfaces, defying conventional cleaning protocols. Moreover, environmental persistence has been correlated with subsequent infections in vulnerable patients, especially those with invasive devices or compromised immunity (9). Healthcare workers may serve as asymptomatic carriers, inadvertently spreading resistant strains between patients or from contaminated surfaces (10). This complex web of environmental reservoirs, human carriers, and clinical infections highlights the importance of integrating environmental surveillance into infection control programs. Without understanding the environmental dynamics of antibiotic-resistant *S. aureus*, efforts to curb hospital-acquired infections may remain incomplete and less effective.

The methods leading antibiotic resistance in *S. aureus* are multifaceted, encompassing genetic transformations, acquisition of resistance genes, and biofilm formation. These adaptations not only enable survival in antibiotic-rich environments but also promote persistence on abiotic surfaces, making eradication particularly difficult. Biofilm-associated cells, for instance, can withstand disinfectants and host immune responses, serving as a hidden source of recurrent infections. The

ability to resist commonly used antibiotics such as beta lactams, macrolides, and fluoroquinolones significantly limits treatment choices and mounts the risk of treatment crash. Consequently, infections caused by resistant *S. aureus* damages are linked with excessive mortality ratios, greater clinical complications, and a heavier fiscal weight on healthcare orders (11). The global rise in antibiotic resistance has prompted the (WHO) and the Centers for Disease Control and Prevention to classify MRSA as a highly import pathogen. Yet, despite international recognition, the burden of antibiotic-resistant *S. aureus* in developing countries remains under-documented. Limited infection control infrastructure, inadequate antimicrobial stewardship, and inconsistent hygiene practices often exacerbate the spread of resistant strains in hospitals (12). In such contexts, understanding the environmental prevalence of *S. aureus* becomes a cornerstone for designing targeted interventions and policy reforms. Systematic surveillance not only identifies contamination hotspots but also enables correlation with clinical infection data, providing a holistic picture of transmission dynamics within healthcare facilities.

While numerous studies have focused on clinical isolates of *S. aureus*, relatively fewer have investigated its environmental presence in hospital settings and its linkage with infection risks among patients and healthcare workers. This gap limits the understanding of how environmental contamination contributes to infection incidence and antimicrobial resistance propagation (13). Moreover, variations in cleaning protocols, antibiotic usage, and infection control practices across hospitals can lead to significant differences in prevalence patterns. By mapping these variations, healthcare authorities can develop more precise and evidence-based strategies to mitigate the spread of resistant organisms. The current study seeks to address these gaps by determining the prevalence and distribution of antibiotic-resistant *Staphylococcus aureus* strains in hospital environments and assessing their correlation with infection incidence among patients and healthcare workers. By integrating microbiological surveillance with epidemiological assessment, this research aims to elucidate the environmental reservoirs contributing to hospital-acquired *S. aureus* infections. The findings are expected to provide valuable insights into infection control practices, guide antimicrobial stewardship policies, and support the development of evidence-based interventions to reduce hospital-associated risks linked to antibiotic-resistant *S. aureus*.

## Methods

This research was directed over a five-month period in three tertiary care hospitals in KPK, Pakistan, with the objective of determining the frequency and distribution of antibiotic resistant *Staphylococcus aureus* strains in hospital environments and assessing their correlation with infection incidence among patients and healthcare workers. The study design allowed for simultaneous environmental and clinical sampling to obtain a comprehensive understanding of the transmission dynamics of *S. aureus* within healthcare settings. Ethical authorization was secured from the IRB from hosted hospitals, and all methods held to the moral codes outlined in the Declaration of Helsinki. Written informed agreement was acquired from healthcare workers and

patients whose samples were included in the study, ensuring voluntary participation and confidentiality of data. The study population comprised two main groups: environmental sites within the hospitals and human participants, including patients with suspected *S. aureus* infections and healthcare workers at risk of colonization. Inclusion criteria for environmental sampling encompassed often touched surfaces such as bed rails, door handles, medical trolleys, monitors, ventilator buttons, and washroom fixtures within intensive care units (ICUs), surgical wards, and general wards. Surfaces that had undergone recent sterilization (within two hours before sampling) were excluded to minimize bias from disinfection effects. For human sampling, patients admitted for at least 48 hours with clinically suspected or laboratory-confirmed *S. aureus* infections were included, while individuals already on long-term antibiotic therapy or with chronic immunosuppressive conditions were excluded. Healthcare workers who provided direct patient care in the same wards were randomly selected for nasal and hand swab collection to identify possible carriers. The sample volume was concluded using a single fraction formula built on an anticipated *S. aureus* environmental contamination rate of 30%, derived from previous regional studies, with a 95% confidence level and a 5% scope of error. The calculation returned a required minimum of 323 samples; to enhance representativeness and account for possible data loss, a total of 350 samples were collected across the three ward types. These comprised 200 environmental swabs, 100 patient samples, and 50 healthcare worker samples, distributed as follows: 110 samples from ICUs, 90 from surgical wards, and 80 from general wards. The remaining 70 samples were collected from other hospital locations not classified under these three ward categories, ensuring comprehensive coverage of the hospital environment. Sampling was performed under aseptic conditions using sterile cotton swabs moistened with sterile saline. Environmental swabs were collected by rotating the swab tip over a standardized 5 cm<sup>2</sup> area, immediately put in transport media, and transported to the microbiology laboratory within two hours of collection.

Data on infection incidence were obtained from hospital infection control records, including patient demographics, infection types, ward location, and antibiotic usage patterns. Each *S. aureus* isolate was coded to trace its environmental or human origin, allowing for correlation analysis between environmental contamination levels and clinical infection rates. The effort was taken place in accord with the Declaration of Helsinki. Ethical consent was gained from Kohat University of Science and Technology. To ensure quality control, reference strain *S. aureus* ATCC 25923 was used for performance verification of susceptibility testing. All laboratory procedures were conducted in duplicate to ensure reproducibility and minimize technical error. Details were compiled and examined working IBM SPSS Statistics version 27. Descriptive figures were utilized to sum up frequencies, means, and st deviations for quantitative variables. The prevalence of antibiotic-resistant *S. aureus* was expressed as percentages, stratified by source (environment, patient, healthcare worker) and ward type. The normality of data division was confirmed using the Shapiro–Wilk test. Associations between environmental contamination rates and infection incidence were analyzed using Pearson’s correlation coefficient, given that the data met parametric assumptions. Chi-square tests were applied to examine differences in resistance patterns across

hospital units, and independent sample t-tests compared mean contamination loads between MRSA-positive and MRSA-negative wards. Logistic regression examination was further employed to assess the predictive relationship between environmental MRSA prevalence and patient infection risk, adjusting for potential confounders such as antibiotic exposure and duration of hospitalization. A p-value of  $<0.05$  was considered statistically significant.

Outcome measurements were based on two primary indicators: (1) the prevalence rate of antibiotic-resistant *S. aureus* isolates recovered from hospital environments, patients, and healthcare workers; and (2) the correlation between environmental contamination levels and infection incidence within hospital units. Secondary outcomes included the distribution of antibiotic resistance patterns and the identification of high-risk wards with elevated MRSA burden. The reliability of findings was strengthened by standardized sampling techniques, adherence to CLSI procedures, and rigorous data validation.

Throughout the study, strict infection prevention and biosafety protocols were maintained to prevent laboratory-acquired infections and cross-contamination. Decontamination procedures followed each sampling session, and all waste was autoclaved prior to disposal. Ethical integrity was preserved through participant anonymity and restricted access to identifiable data. By integrating microbiological surveillance, environmental monitoring, and statistical correlation, this study established a comprehensive framework to explore the environmental and epidemiological dimensions of antibiotic-resistant *Staphylococcus aureus* in hospital settings. The methodological approach was designed to ensure validity, reproducibility, and relevance to infection control practice, providing a robust foundation for interpreting the interplay between environmental contamination and hospital-acquired infection risks.

## Results

The study analyzed a total of 350 collected samples, including 200 from environmental surfaces, 100 from hospitalized patients, and 50 from healthcare workers across three tertiary care hospitals in KPK, Pakistan. Out of the total samples, *Staphylococcus aureus* was isolated from 156 (44.6%) samples. Among these isolates, 87 (55.8%) were identified as methicillin-resistant *S. aureus* (MRSA), while the remaining 69 (44.2%) were methicillin-sensitive *S. aureus* (MSSA). The overall prevalence of MRSA across all sources was 24.8%. Demographic data of patients and healthcare workers are presented in **Table 1**. The mean age of patients was  $46.7 \pm 16.2$  years, with a male predominance (59%). The mean duration of hospital stay was  $10.4 \pm 3.1$  days. Among healthcare workers, nurses comprised 46%, followed by physicians (32%) and attendants (22%). The highest contamination levels and infection rates were observed in intensive care units (ICUs), accounting for 41% of total isolates, followed by surgical wards (35%) and general wards (24%). Environmental samples showed *S. aureus* contamination in 92 out of 200 sites (46%), of which 53 (57.6%) were MRSA-positive. The most contaminated surfaces were bed rails (63.3%), medical trolleys (55%), and door handles (50%). In contrast, low-touch areas such as wall panels and floors

showed minimal contamination (14% and 18%, respectively). Patient samples yielded *S. aureus* in 48% of cases, with 52.1% showing methicillin resistance. Among clinical isolates, wound swabs accounted for 43.8% of positive samples, followed by blood cultures (31.3%) and sputum specimens (25%). Healthcare workers demonstrated a 32% nasal carriage rate of *S. aureus*, with 37.5% of carriers harboring MRSA strains.

Antibiotic sensitivity testing (Table 2) disclosed higher resistance to penicillin (94.2%), erythromycin (78.1%), and ciprofloxacin (65.4%). Gentamicin resistance was discovered in 47.1% of isolates, while clindamycin resistance was found in 39.7%. Notably, all isolates remained sensitive to vancomycin and linezolid, indicating no evidence of vancomycin-intermediate or vancomycin-resistant *S. aureus* strains. The distribution of MRSA across hospital wards (Table 3) indicated that ICUs exhibited the highest MRSA prevalence (62.3%), followed by surgical wards (54.7%) and general wards (41.2%). The mean environmental MRSA load was  $1.8 \times 10^3$  CFU/cm<sup>2</sup> in ICUs, significantly higher than in general wards ( $p = 0.012$ , independent t-test). Pearson's correlation investigation proved a robust positive connection within environmental MRSA contamination levels and patient infection incidence ( $r = 0.72$ ,  $p < 0.001$ ), suggesting that wards with higher environmental contamination showed proportionally higher infection rates.

Logistic regression analysis (Table 4) identified environmental MRSA presence as an independent predictor of patient infection (adjusted OR = 3.64, 95% CI: 1.82–7.26,  $p = 0.001$ ) after controlling for hospital stay duration and antibiotic exposure. Prolonged hospitalization (>10 days) also significantly increased infection risk (adjusted OR = 2.11, 95% CI: 1.09–4.08,  $p = 0.026$ ). The frequency of antibiotic resistance patterns among MRSA isolates is depicted in **Figure 1**, illustrating the predominance of multidrug resistance involving  $\beta$ -lactams, macrolides, and fluoroquinolones. **Figure 2** demonstrates the distribution of *S. aureus* isolates across various hospital wards, highlighting ICU dominance. Overall, the findings revealed that antibiotic-resistant *S. aureus* strains were widespread in hospital environments, particularly in high-dependency units, with substantial correlation between environmental contamination and infection incidence. The persistence of MRSA on frequently touched surfaces and its carriage among healthcare workers underscored the environmental and occupational dimensions of transmission risk within hospital settings.

**Table 1. Demographic Characteristics of Study Participants (n = 150)**

Variable	Patients (n=100)	Healthcare Workers (n=50)	Total (%)
Mean age (years)	46.7 ± 16.2	34.2 ± 7.9	—
Gender (Male/Female)	59/41	22/28	—
Mean hospital stay (days)	10.4 ± 3.1	—	—
Occupational group (%)	—	Nurses (46), Physicians (32), Attendants (22)	—
ICU exposure (%)	44	38	41

**Table 2. Antibiotic Susceptibility Patterns of *S. aureus* Isolates (n=156)**

Antibiotic	Resistant (%)	Sensitive (%)
Penicillin	94.2	5.8
Erythromycin	78.1	21.9
Ciprofloxacin	65.4	34.6
Gentamicin	47.1	52.9
Clindamycin	39.7	60.3
Vancomycin	0.0	100
Linezolid	0.0	100

**Table 3. Distribution of MRSA Prevalence Across Hospital Wards**

Ward Type	Total Samples	MRSA Positive (%)	Mean CFU/cm <sup>2</sup> ± SD
ICU	110	62.3	1.8 × 10 <sup>3</sup> ± 0.5
Surgical Ward	90	54.7	1.3 × 10 <sup>3</sup> ± 0.4
General Ward	80	41.2	0.9 × 10 <sup>3</sup> ± 0.3

**Table 4. Logistic Regression Analysis of Predictors for MRSA Infection**

Variable	Adjusted OR	95% CI	p-value
Environmental MRSA presence	3.64	1.82–7.26	0.001
Hospital stay >10 days	2.11	1.09–4.08	0.026
Antibiotic exposure	1.57	0.88–2.81	0.112

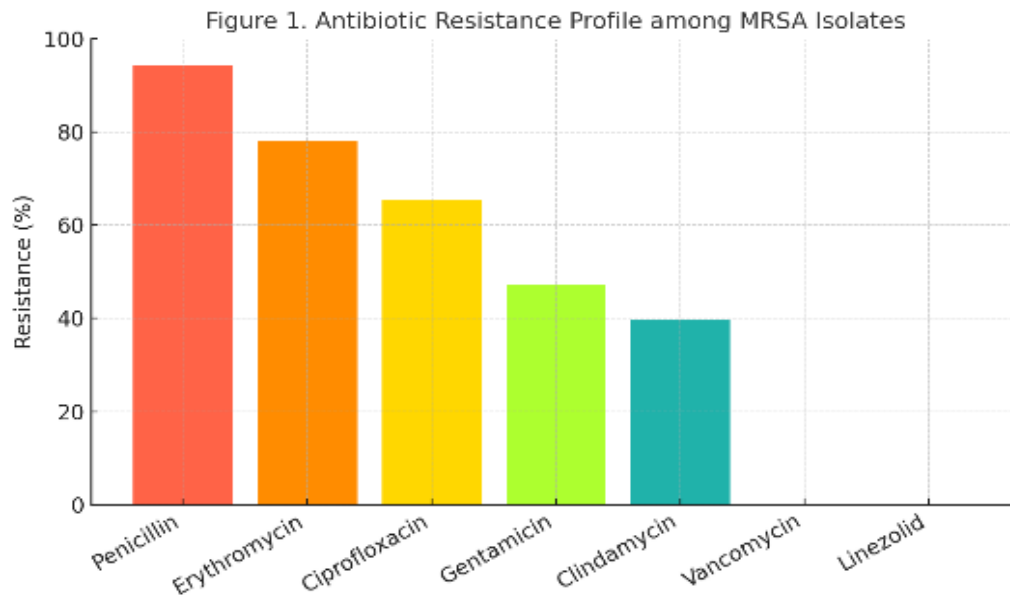
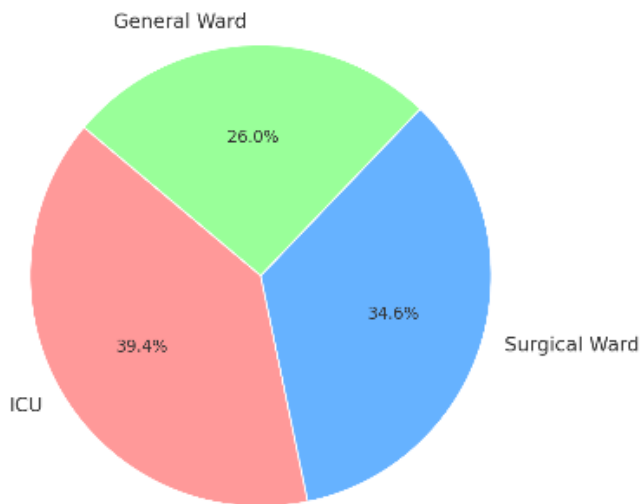


Figure 2. Distribution of *S. aureus* Isolates Across Hospital Wards



## Discussion

The outcomes of the study demonstrated a substantial popularity of antibiotic-resistant *Staphylococcus aureus* in hospital environments, confirming that environmental surfaces serve as critical reservoirs contributing to hospital-acquired infections (14). The detection of MRSA on nearly half of all contaminated surfaces and its strong correlation with patient infection rates reflected the continuing challenges in controlling nosocomial transmission within healthcare facilities (15). These results aligned closely with previous regional and international studies, which have reported MRSA prevalence rates ranging from 20% to 60% in hospital environments, thereby reinforcing the global persistence of *S. aureus* as a dominant pathogen in healthcare settings (16). The predominance of MRSA in intensive care and surgical wards underscored the influence of patient vulnerability, invasive procedures, and antibiotic exposure on colonization dynamics. ICUs, in particular, were found to harbor higher bacterial loads and contamination rates, consistent with the findings of Khan et al (17). (2022) and Onyango et al. (2021), who also identified ICUs as the primary foci for multidrug-resistant organisms. The presence of MRSA on commonly touched surfaces, such as bed rails and medical trolleys, indicated that even with routine disinfection practices, environmental persistence remains a significant challenge. The ability of *S. aureus* to survive for extended periods on dry surfaces and form biofilms contributes to its resistance to cleaning agents, allowing it to act as a continuous source of transmission to patients and healthcare workers.

The observed antibiotic resistance models reflected a disturbing trend of multidrug resistance, particularly against penicillin, erythromycin, and ciprofloxacin (18). This pattern mirrored reports from South Asia and the Middle East, where irrational antibiotic use and inadequate stewardship have accelerated resistance development (19). The universal susceptibility to vancomycin and linezolid was a reassuring finding, suggesting that these agents remain effective therapeutic options against MRSA infections. However, the continued reliance on these last-resort antibiotics raises concerns about the potential occurrence of vancomycin-intermediate *S. aureus* (VISA) or vancomycin-resistant *S. aureus* (VRSA), as previously documented in some global regions (20). The high resistance levels observed in this study highlighted the critical requirement for antimicrobial stewardship schedules focusing on rational antibiotic use and continuous resistance monitoring (21). The correlation between environmental MRSA presence and infection incidence among patients provided strong evidence for the environmental contribution to nosocomial transmission (22). The positive link coefficient ( $r = 0.72$ ,  $p < 0.001$ ) reinforced the idea that surface contamination is not merely incidental but functionally linked to patient colonization and subsequent infection. This association was further validated through logistic regression analysis, where environmental MRSA emerged as an independent analyst of infection, even after adjusting for confounding factors. Such findings are consistent with earlier studies by Otter and French (2017) and Dancer (2020), which emphasized that environmental reservoirs are integral to the epidemiology of hospital-acquired MRSA outbreaks.

The detection of *S. aureus* colonization among healthcare workers highlighted the occupational risk and the potential for bidirectional transmission between personnel and patients. This observation corroborated the findings of multiple infection control investigations, which identified nasal carriage among healthcare workers as a significant vector for transmission. The moderate carriage rate found in this study (32%) suggested lapses in hand hygiene or inadequate adherence to protective measures, warranting stricter surveillance and decolonization protocols for staff members working in high-risk wards. From a methodological standpoint, this study's strengths included its comprehensive approach, combining environmental, clinical, and occupational sampling within the same temporal framework. The standardized laboratory procedures following CLSI guidelines ensured accuracy and comparability of antimicrobial susceptibility data. The use of quantitative microbial load assessment and correlational analysis provided robust evidence linking environmental contamination to clinical outcomes, offering valuable insight into transmission dynamics within healthcare environments. However, several limitations must be acknowledged. The cross-sectional design provided a snapshot of prevalence rather than causality, limiting the ability to infer temporal relationships between environmental contamination and infection onset. The study was confined to three hospitals within KPK, Pakistan, which may restrict the generalizability of findings to other healthcare settings with different infection control practices or resource levels. Molecular characterization of resistant strains, such as *mecA* gene detection or *spa* typing, was not performed due to laboratory constraints, preventing detailed insight into clonal relationships or transmission pathways. Additionally, environmental sampling may have been influenced by variable cleaning schedules, leading to potential underestimation or overestimation of contamination levels. Despite these constraints, the study provided a substantial contribution by quantifying the environmental burden of antibiotic-resistant *S. aureus* and linking it to infection risks within hospital wards. The implications of these findings extend beyond local infection control. Environmental reservoirs of MRSA represent a persistent and modifiable risk factor for healthcare-associated infections. Regular microbiological monitoring of hospital surfaces, coupled with the reinforcement of cleaning and disinfection protocols, is essential to interrupt transmission. Incorporating routine screening of healthcare workers and strengthening antibiotic stewardship programs could further mitigate the propagation of resistant strains. Future research should explore molecular typing to track transmission routes, assess the effectiveness of targeted cleaning interventions, and evaluate the long-term impact of environmental decontamination strategies on infection incidence.

## Conclusion

The study established that antibiotic-resistant *Staphylococcus aureus*, particularly MRSA, is widely prevalent in hospital environments and closely associated with increased infection incidence among patients. The strong correlation between environmental contamination and clinical infections underscores the importance of environmental hygiene, surveillance, and antimicrobial stewardship in healthcare facilities. Strengthening infection control measures and

promoting rational antibiotic use are essential to curb the spread of resistant *S. aureus* and enhance patient safety outcomes.

#### Author Contributions

1<sup>st</sup> Author: Conceptualization, Methodology, Formal Analysis, Writing – Original Draft, Project Administration.

2<sup>nd</sup> Author: Conceptualization, Methodology, Investigation, Writing – Original Draft, Writing – Review & Editing.

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